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# Efficacy of preplant soil fumigation with chloropicrin for tomato production in Italy

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#### Abstract

The efficacy of chloropicrin (CP) as a possible alternative fumigant to methyl bromide (MB) was investigated in tomato production systems in Northern (Albenga, Liguria) and Southern (Acate, Sicily) Italy, in six experimental trials. Two different application methods were tested: soil injection using traditional shank fumigation equipment and drip fumigation through the irrigation systems using different amounts of water to deliver CP. In the presence of severe attacks of *Verticillium dahliae*, *Fusarium oxysporum* f. sp. *lycopersici* and *Fusarium oxysporum* f. sp. *radicis-lycopersici*, CP applied by shank injection at rates  $\geq 30 \text{ g/m}^2$ , provided a satisfactory and consistent control of tomato diseases, without causing phytotoxicity. However, CP applied by shank injection at 30 or  $40 \text{ g/m}^2$  was less effective than MB applied using the standard rate ( $60 \text{ g/m}^2$ ) and method. The concentration of CP providing the most control of fungal pathogens ranged from 400 to  $700 \,\mu\text{l/l}$ . CP efficacy applied through the drip irrigation systems seemed affected by the amount of irrigation water more than the application rate or concentration. Soil type and organic matter content may influence the efficacy of the treatment, particularly when CP is applied through shank injection. With this type of application, the same application rate of CP was more effective in Acate soil that contains more sand and less organic matter than in Albenga soil. For any application rate of CP, the efficacy of drip fumigation was greater than shank injection, particularly in the Albenga soil. © 2002 Elsevier Science Ltd. All rights reserved.

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# 1. Introduction

The tomato (Lycopersicon esculentum Miller) is a high-value crop in Italy, grown in greenhouses or in fields, and subject to great losses caused by several soil pests such as fungal pathogens and nematodes. The most important fungal pathogens that can devastate tomato crops are Fusarium oxysporum f. sp. lycopersici, F. oxysporum f.sp. radicis-lycopersici, and Verticillium dahliae. Methyl bromide (MB) fumigation has provided a reliable and effective treatment to control pests and diseases in tomato crops in Italy, as well as all over the world, since the 1960s. Tomato crops account for about 30% of the consumption of MB in the world (UNEP,

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1995). Recently, however, MB has been implicated in the destruction of the ozone layer (Bell et al., 1996; Duniway et al., 2000; Garibaldi and Gullino, 1995; Mellouki et al., 1992; Ristaino and Thomas, 1998; Solomon and Albritton, 1992) and included among the substances with high ozone-depleting potentials (ODP) by the Montreal Protocol, an international treaty sponsored by the United Nations Environment Programme (UNEP). This treaty calls for a phase-out of MB by 2005. With the impending loss of MB, research is currently under way to develop alternative crop production practices and to find reliable alternatives to this fumigant.

In the case of tomatoes, soil solarization could partially replace chemical fumigation in certain areas (mostly Central and Southern Italy) and in certain production systems. Also, the use of resistant cultivars and of grafting can reduce the pressure of soil-borne diseases. However, in the case of high-value fresh market tomato crops, soil fumigation remains essential

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in many production systems. Of the currently available fumigants, chloropicrin (CP) (trichloronitromethane, CCl<sub>3</sub>NO<sub>2</sub>) is the most efficacious against plant pathogenic fungi (Locascio et al., 1997; Nadakavukaren and Horner, 1959). Historically, CP, not yet registered in Italy, has been used in combination with MB at different ratios and for different purposes. In formulations containing 98% MB and 2% CP, the added CP serves only as an efficient warning agent. Other mixtures (the most common is 66/33 MB/CP) have been developed and applied in many countries to broaden the spectrum of activity and enhance the control of soil-borne fungal pathogens (Wilhelm et al., 1961). Although it is well known that the combinations of MB and CP act synergistically against soil-borne fungi (Wilhelm, 1966), the biological activity of CP is not fully known, but it is believed that CP reacts with biological thiols disrupting multiple targets within the cell (Sparks et al., 1997).

Recent studies of alternative fumigants to MB for tomato production in Florida found that CP shank injected at 390 kg/ha provided good control of nematodes and soil fungi (Locascio et al., 1997). However, tomato yield was significantly lower in the CP fumigation than yields following from MB fumigation. Although some information is available on the efficacy of MB–CP mixtures to control soil-borne pathogens and weeds, little is known about the efficacy of CP alone for tomato production under various soil and environmental conditions. Also, little information is available on the efficacy of CP applied with water through drip irrigation systems.

In our study, the efficacy of CP as a possible alternative fumigant to MD was investigated in tomato production systems in two Italian geographical areas. Two different application methods were evaluated: soil injection using traditional shank fumigation equipment and drip fumigation through the irrigation systems using different amounts of water to deliver CP.

#### 2. Materials and methods

#### 2.1. Study sites

Six experimental trials were carried out in 1999 and 2000 in two geographical areas in Italy. One site was located in Southern Italy (Acate, RG, Sicily) at the commercial farm "S.I.S. Mogli". The other site was located in Northern Italy (Albenga, SV, Liguria) at the Experimental Station of the Chamber of Commerce of Savona. Each experimental site had a history of at least one tomato crop during the three seasons prior to the beginning of this study. In Sicily, during the previous year's tomato crop, severe losses due to Pyrenochaeta lycopersici and Fusarium oxysporum f.sp. lycopersici were reported by the local growers' extension service. At the Acate location, two trials were carried out in the open field on a loamy sand soil (sand, 82%; silt, 7%; clay, 11%; pH, 8.3; organic matter content, 0.7%; and cation exchange capacity, 5.0 meg/100 g soil). Both trials were carried out in the same field. In the interval between the trials, the soil was deeply rototilled and mixed. At the Albenga location, four trials were carried out on a sandy loam (sand, 75%; silt, 20%; clay, 5%; pH, 8.1; organic matter content, 2.5%; and cation exchange capacity, 8.5 meg/100 g soil). Two trials took place in the open field and two in greenhouses. A randomized complete block design was used with four replicates. The layout of the six trials is summarized in Table 1. Tomato seedlings were transplanted into preformed beds (80 cm wide, 30 m long, and 10 cm high, 24 m<sup>2</sup>) which were immediately covered after shank fumigation with a plastic mulch. Black polyethylene film (0.05 mm thickness) was used (Eiffel, Fontanellato, Parma, Italy) to cover the soil beds at the time of treatment and buried at 20 cm depth. The nontreated control beds were also covered with this film. Each bed represented one plot and was located 80 cm from other parallel beds (plots). Each bed was irrigated with one

Table 1 Experimental layout of the six trials

Trial	Site	Type	CP rate (g/m <sup>2</sup> )	Application method	Fumigation	Mulch holing	Irrigation	Tomato transplant	End of the trial
1	Acate	Open field	20, 30, 40	Shank injection	June 30, 1999	July 21, 1999	July 28, 1999	August 5, 1999	December 1999
2	Albenga	Open field	20, 40	Shank injection	July 8, 1999	July 26, 1999	July 28, 1999	August 2, 1999	December 1999
3	Albenga	Open field	20, 40	Drip irrigation	July 8, 1999	July 26, 1999	July 23, 1999	August 2, 1999	December 1999
4	Albenga	Greenhouse	20, 40	Drip irrigation	August 10, 1999	August 23, 1999	August 24, 1999	August 31, 1999	December 1999
5	Acate	Open field	20, 40, 60	Drip irrigation	March 28, 2000	April 19, 2000	April 20, 2000	May 18, 2000	September 2000
6	Albenga	Greenhouse	20, 40, 60	Drip irrigation	April 20, 2000	May 10, 2000	May 11, 2000	May 16, 2000	September 2000

drip line placed under the plastic film. Plots were fertilized at 10 day intervals with N:P:K 20:10:10 during the cultivation period. The fertilizer was provided by means of the drip irrigation system in five applications (100 kg/ha each) according to the local cultural practices. At least 2 weeks after fumigation, the soil was kept mulched and holes were then cut in the plastic mulch and the soil was irrigated with at least 40 mm of well water (Table 1).

## 2.2. Soil infestation with pathogens

To achieve a uniform soil infestation and higher disease pressure before fumigation, an artificial inoculum of Fusarium oxysporum f.sp. lycopersici was incorporated into the soil. Two strains of the pathogen, freshly isolated from infected tomato plants, were grown on autoclaved wheat kernels. Twenty g/m<sup>2</sup> of infested kernels were incorporated into the soil by rototilling at a depth of 10-15 cm 7-10 days prior to fumigation. Soil was kept moist for 2 weeks by means of periodic sprinkler irrigation (5-10 mm). In addition, the effect of fumigation on the survival of selected fungi was evaluated through a bioassay of bags containing several pathogens buried in the soil. The pathogens tested (Fusarium oxysporum f.sp. lycopersici, Fusarium oxysporum f.sp. melonis, Rhizoctonia solani, Sclerotinia sclerotiorum and Verticillium dahliae) were introduced as infected wheat kernels. A 7-g inoculum of each pathogen was placed separately into small gas permeable tissue bags (5 cm diam) and buried in the soil 12-18 h before the fumigation. Two bags per pathogen were placed in the center of each plot, one at 10 and one at 20 cm depth. The bags were extracted 14 days after fumigation when holes were punched in the mulch before soil watering.

# 2.3. Pathogen inoculum survival (bioassay)

The bags containing the inocula were taken out of the soil and immediately transferred to the laboratory in a portable refrigerator (8–10°C), and processed. One hundred infected kernels/bag were placed on Petri plates (10 kernels/plate) containing potato dextrose agar (PDA) or a semi-selective media for *Fusarium* (Komada, 1975), *Rhizoctonia* (Migheli et al., 1990), or *Verticillium* (Nadakavukaren and Horner, 1959). Plates inoculated with *F. oxysporum*, *R. solani* and *S. sclerotiorum* were incubated at 23–24°C (12 h photoperiod), while plates inoculated with *V. dahliae* were incubated at 17–18°C in the dark. The number of kernels with growing mycelium was counted, and the data are expressed as percentage of infected kernels.

# 2.4. Chloropicrin application

A commercial formulation of CP (Tripicrin, 99% a.i., 1.65 kg/l density, Trical, Hollister, CA, USA) was used throughout the work. The fumigant was applied by soil injection or by drip irrigation at application rates reported in Tables 2–12.

In trials 1 and 2, CP was mechanically applied with the same equipment used for MB cold injection. In a single operation, this equipment is able to form beds (80 cm width and 10 cm high), install the drip lines, and cover the soil with plastic mulch during fumigant injection. In each bed, CP was injected at 30 cm depth with two shanks placed 20 cm from the bed edge and 40 cm apart.

The application of CP by drip irrigation (trials 3–6) was carried out on beds, mechanically made with the same equipment employed for injection. Each bed was provided with two drip lines, placed on the soil surface, 20 cm from the bed edge and 40 cm from each other. The

Table 2
Effect of fumigation with chloropicrin (CP), applied by soil injection, and methyl bromide (MB) on the number of tomato plants [Principe Borghese (PB) and Vulcano (V)] infested with *P. lycopersici, F. oxysporum* f.sp. *radicis lycopersici, F. oxysporum* f.sp. *lycopersici* and *V. dahliae* (Acate, trial 1, 1999)

Application rate (g/m <sup>2</sup> )	Percentag	Combine	d					
	P. lycopersici		Wilt pathogens <sup>a</sup>		F. oxysporum f.sp. radicis lycopersici			
	PB	V	PB	V	PB	V	PB	V
Control, -	9.9b <sup>b</sup>	14.9c	4.3b	5.3a	22.5b	25.5b	36.7b	45.7b
CP, 20	0.0a	0.0a	0.5a	2.7a	5.9a	3.5a	6.4a	6.2a
CP, 30	1.6a	0.0a	0.5a	1.7a	5.2a	7.5a	7.3a	9.2a
CP, 40	4.3ab	4.9b	0.0a	1.5a	3.2a	2.1a	7.5a	8.5a
MB, 60	1.1a	0.6a	0.0a	0.6a	0.5a	1.8a	1.6a	3.0a

<sup>&</sup>lt;sup>a</sup>The value includes the percent of plants infected by *V. dahliae* and by *F. oxysporum* f.sp. *lycopersici*.

<sup>&</sup>lt;sup>b</sup> Means of the same column followed by the same letter do not differ according to Duncan's Multiple Range Test (P = 0.05).

drip lines (17 mm diameter) were equipped with water emitters (flow rate 2.4 l/h) spaced 30 cm apart. The commercial formulation of CP was injected into the

Table 3
Effect of fumigation with chloropicrin (CP), applied by soil injection, and methyl bromide (MB) on the yield of tomato plants [Principe Borghese (PB) and Vulcano (V)] (Acate, trial 1, 1999)

Application rate (g/m <sup>2</sup> )	Fruit size (g/fruit)		Yield (Kg/m	<sup>2</sup> )	No. fruits/m <sup>2</sup>		
	PB	V	PB	V	PB	V	
Control, -	150a <sup>a</sup>	694a	2.1b	3.6b	84b	28b	
CP, 20	156a	695a	3.4a	6.7a	127a	49a	
CP, 30	148a	720a	3.3a	7.3a	128a	52a	
CP, 40	143a	704a	3.3a	7.1a	125a	58a	
MB, 60	159a	714a	3.7a	7.5a	134a	54a	

<sup>&</sup>lt;sup>a</sup> Means of the same column followed by the same letter do not differ according to Duncan's Multiple Range Test (P = 0.05).

Table 4
Effect of fumigation with chloropicrin (CP), applied by soil injection, and methyl bromide (MB) on the number of tomato plants [cv Principe Borghese (PB)] infested with *V. dahliae*, *F. oxysporum* f.sp. *lycopersici* and *F. oxysporum* f.sp. *radicis lycopersici* (Albenga, trial 2, 1999)

Application rate (g/m²)	Control on									
(2)	27/09/99			28/10/99						
	% plants in	fected by	Total	% plants in	fected by	Total				
	Wilt pathogens <sup>a</sup>		\ /		F. radicis lycopersici	(%)				
Control, - CP, 20 CP, 40 MB, 60	5.7a <sup>b</sup> 6.8a 7.2a 5.0a	17.9b 16.7ab 18.2b 14.0a	23.5b 25.4b	25.6b 19.5ab 17.1ab 11.7a	29.6b 27.2a 30.7b 27.5a	55.2c 46.7b 47.8b 39.2a				

<sup>&</sup>lt;sup>a</sup> The value includes the percent of plants infected by *V. dahliae* and *F. oxysporum* f.sp. *lycopersici*.

irrigation line by a peristaltic pump, adjusted to the required final concentration. CP was applied with different irrigation water amounts, ranging from 17 to 52 mm. This corresponds to different concentrations of the fumigant, varying from 400 to 1200  $\mu$ l/l. Since CP is only slightly soluble in water (1600 mg/l), an emulsifier (TS101, Trical, Hollister, CA, USA) was used (5% by wt) in all applications to ensure uniform distribution of CP in the irrigation water.

All trials included an untreated control and methyl bromide (60 g/m², hot gas application) treated plots. The application rate of MB corresponds to the rate applied under standard polyethylene film. Twenty days after fumigation, and after removing the inoculum bags, the soil was post-irrigated with at least 50 mm of water.

## 2.5. Tomato transplant and cultural practices

Forty tomato plants, 50 days old, belonging to the cultivar Principe Borghese (cherry-type, with a single fruit harvest, SAIS, Cesena, Forlì, Itali) and 40 plants belonging to the cultivar Vulcano (tomato in bunches of 3–4 fruits Olter Sementi, Asti, Italy) were transplanted 7 days after the post-irrigation, on the dates reported in Table 1. Plants were placed in two rows per bed, approximately 15–20 cm from the bed edge and 40–50 cm apart, and at 40 cm spacing along each row. Plants were drip irrigated and grown according to the cultural practices adopted by local commercial growers. From the transplant date, at least eight sprays with insecticides aimed at reducing the presence of virus vectors were applied at 7–10 day intervals.

## 2.6. Data collection and analysis

The disease development was evaluated at regular intervals by counting the number of plants with symptoms (disease pressure); the infected plants were counted and eliminated. Also, the number of healthy plants was counted. The yield was evaluated on the

Table 5
Effect of fumigation with chloropicrin (CP), applied by drip irrigation, and methyl bromide (MB) on tomato plants [Principe Borghese (PB)] infested with *V. dahliae*, *F. oxysporum* f.sp. *lycopersici* and *F. oxysporum* f.sp. *radicis lycopersici* (Albenga, trial 3, 1999)

Application rate (g/m <sup>2</sup> ), mm water (CP μl/l)	Control on										
иши жиег (Ст <b>р</b> ідт)	08/09/99		28/10/99								
	% plants infected	by	Total	% plants infected	Total						
	Wilt pathogens <sup>a</sup>	F.radicis lycopersici	(%)	Wilt pathogens <sup>a</sup>	F.radicis lycopersici	(%)					
Control, -, -	3.3a <sup>b</sup>	17.5b	20.8b	25.1b	41.0b	66.1c					
CP, 20, 17 (700)	0.6a	8.3ab	8.9a	12.2a	38.1ab	50.3b					
CP, 40, 20 (1,200)	2.0a	11.5ab	13.5ab	10.3a	29.4ab	39.7ab					
MB, 60, -	1.9a	4.6a	6.5a	8.5a	28.5a	37.0a					

<sup>&</sup>lt;sup>a</sup>The value includes the percent of plants infected by *V. dahliae* and by *F. oxysporum* f.sp. *lycopersici*.

<sup>&</sup>lt;sup>b</sup>Means of the same column followed by the same letter do not differ according to Duncan's Multiple Range Test (P = 0.05).

<sup>&</sup>lt;sup>b</sup> Means of the same column followed by the same letter do not differ according to Duncan's Multiple Range Test (P = 0.05).

Table 6
Effect of fumigation with chloropicrin (CP), applied by drip irrigation, and methyl bromide (MB) on the yield of tomato plants (Principe Borghese) (Albenga, trial 3, 1999)

Application rate (g/m²), mm water (CP μl/l)	Kg/m <sup>2</sup>	No. fruits/m <sup>2</sup>	g/fruit
Control, -, -	0.3b <sup>a</sup>	17.8b	21.6a
CP, 20, 17 (700)	0.4ab	23.8ab	18.7a
CP, 40, 20 (1,200)	0.6ab	25.4ab	21.8a
MB, 60, -	0.7a	31.3a	22.5a

<sup>&</sup>lt;sup>a</sup> Means of the same column followed by the same letter do not differ according to Duncan's Multiple Range Test (P = 0.05).

Table 7
Effect of fumigation with chloropicrin (CP), applied by drip irrigation, and methyl bromide (MB) on the number of tomato plants [Principe Borghese (PB) and Vulcano (V)] infested with *V. dahliae* and *F. oxysporum* f.sp. *lycopersici* (Albenga, trial 4, 1999)

Application rate g/m <sup>2</sup> , mm water (CP µl/l)	Percenta	Total (	%)				
. , , ,	Wilt pat	hogens <sup>a</sup>	F. radi lycoper				
	PB	V	PB	V	PB	V	
Control, -, - CP, 20, 20 (600) CP, 40, 35 (700) MB, 60, -		20.2b 14.3ab 9.5a 8.4a	43.0a 25.7a 21.4a 18.6a	65.6c 56.0bc 47.9ab 37.3a	65.9b 38.6ab 27.3a 27.2a	85.8c 70.3b 57.4a 45.7a	

<sup>&</sup>lt;sup>a</sup>The value includes the percent of plants infected by *V. dahliae* and by *F. oxysporum* f.sp. *lycopersici*.

Means of the same column followed by the same letter do not differ

Table 8
Effect of fumigation with chloropicrin (CP), applied by drip irrigation, and methyl bromide (MB) on the yield of tomato plants [Principe Borghese (PB) and Vulcano (V)] (Albenga, trial 4, 1999)

Application rate g/m <sup>2</sup> , mm water (CP µl/l)	g/frui	g/fruit		$Kg/m^2 \\$		No. fruits/m <sup>2</sup>	
mm water (er pa/i)	PB	V	PB	V	PB	V	
Control, -, -	43a <sup>a</sup>	241a	0.2b	0.2b	12.0b	1.6d	
CP, 20, 20 (600)	44a	219a	0.5a	0.3ab	23.1a	3.6c	
CP, 40, 35 (700)	45a	194a	0.5a	0.4ab	23.5a	4.0b	
MB, 60, -	49a	284a	0.6a	0.5a	25.1a	4.9a	

<sup>&</sup>lt;sup>a</sup> Means of the same column followed by the same letter do not differ according to Duncan's Multiple Range Test (P = 0.05).

healthy plants by counting the number of fruits per plant and by weighing the fruits. Data are expressed as number and weight of marketable fruit per plant as well as number of fruits and weight/m². All data collected were statistically analyzed, according to Duncan's Multiple Range Test. In some trials, plants suffered severe infestation with tomato spotted wilt virus (TSWV). Particularly, in trials 2 and 3 the cv Vulcano was highly infested with TSWV. For this reason, in the case of trials 2 and 3, only the results obtained on the cv

Principe Borghese are reported. In the case of the trials carried out in Sicily, tomato yellow leaf curl virus (TYLCV) caused some losses.

#### 3. Results

In five out of six trials, the damage caused by soilborne fungal pathogens was severe. The results are reported according to the different trials.

## 3.1. Trial 1 (Acate, field shank injection)

In this trial, the corky root caused by Pyrenochaeta lycopersici developed early, approximately 100 days after transplanting, and affected 15% of the Vulcano variety plants in the control plots (Table 2). This cultivar appeared more susceptible than "Principe Borghese". Corky root symptoms at a lower infestation level were also observed in plots treated with  $40 \,\mathrm{g/m^2}$  of CP. Fusarium crown and root rot was severe in the control plots on both cultivars, while a few plants exhibited wilts caused by F. lycopersici and Verticillium dahliae. All treatments performed equally well at the end of the trial and all showed less damage than the untreated control (Table 2). All CP and MB treatments improved the number and weight of fruit but not fruit size (Table 3). Plants suffered from infection by TYLCV, which caused serious crop losses. Plants showing symptoms of TYLCV were constantly eliminated before the 2nd bunch emission and were not considered during the controls.

#### 3.2. Trial 2 (Albenga, field shank injection)

Chloropicrin shank injected at both rates did not control crown and root rot of tomato, and only partially reduced wilt pressure (Table 4). Methyl bromide provided a better, although not complete, control of all pathogens. Also CP at 20 and  $40\,\mathrm{g/m^2}$  significantly reduced disease pressure (Table 4). No differences in yield were observed (data not shown), probably due to the high level of TSWV infestation. Plants showing symptoms of TSWV were constantly eliminated before the 2nd bunch emission and were not considered during the controls. The cv Vulcano, heavily damaged, was not considered for disease incidence and production.

## 3.3. Trial 3 (Albenga, field drip fumigation)

In this field trial, crown and root rot and wilts caused very high losses in the control plots (Table 5). All tested treatments significantly reduced wilt symptoms, with methyl bromide providing the best results, particularly in the presence of severe crown and root rot. When the total disease pressure is evaluated, the best results are

Table 9
Effect of fumigation with chloropicrin (CP), applied by drip irrigation, and methyl bromide on the survival of buried soil pathogens (Acate, trial 5, 2000)

Application rate g/m <sup>2</sup> , mm water (CP µl/l)	Percentage of kernels infested at the end of fumigation with											
mm water (CF μi/i)	F. lycopersici		F. melonis		R. solani		S. sclerotiorum		V. dahliae			
Depth (cm)	10	20	10	20	10	20	10	20	10	20		
Control,-, -	100.0b <sup>a</sup>	100.0b	100.0b	100.0b	42.8b	28.3b	56.3b	73.3c	71.0d	90.0d		
CP, 20, 20 (600)	100.0b	83.3ab	45.5a	43.5a	7.5a	18.8ab	32.3a	63.8c	65.3cd	62.7c		
CP, 20, 30 (400)	79.8b	100.0b	53.5a	53.5ab	5.3a	6.0a	16.5a	44.3bc	53.3bc	46.3bc		
CP, 40, 35 (700)	75.8b	82.0ab	42.3a	48.0ab	4.5a	10.0a	9.0a	17.3ab	40.0b	38.7abc		
CP, 60, 52 (700)	37.8a	59.3a	37.3a	41.8a	3.3a	9.0a	14.0a	8.5a	23.3a	30.0ab		
CP, 60, 35 (1,000)	24.8a	53.8a	38.8a	53.8ab	3.5a	4.0a	14.0a	32.8b	9.3a	14.3a		
MB, 60, -	97.5b	84.8ab	46.5a	70.5ab	0.8a	7.8a	21.8a	4.5a	42.0b	24.3ab		

<sup>&</sup>lt;sup>a</sup> Means of the same column followed by the same letter do not differ according to Duncan's Multiple Range Test (P=0.05).

Table 10 Effect of fumigation with chloropicrin (CP), applied by drip irrigation, and methyl bromide (MB) on the number of infested tomato plants [Principe Borghese (PB) and Vulcano (V)] with *V. dahliae, F. oxysporum* f.sp. *lycopersici* and *F. oxysporum* f.sp. *radicis lycopersici* (Albenga, trial 6, greenhouse, 2000)

Application rate g/m <sup>2</sup> , mm water (CP µl/l)	% infected plants										
water (C1 pa/1)	Wilt pathogo	ens <sup>a</sup>	F. radicis lyco	persici	Total % diseased plants						
	PB	V	PB	V	PB	V					
Control,-, -	5.6b <sup>b</sup>	2.5b	26.0c	2.5a	31.6c	5.0a					
CP, 20, 20 (600)	3.1ab	0.0a	19.4b	0.0a	22.5b	0.0a					
CP, 20, 30 (400)	0.0a	2.5b	9.3a	2.5a	9.3a	5.0a					
CP, 40, 35 (700)	0.0a	0.0a	18.9b	0.0a	18.9b	0.0a					
CP, 60, 52 (700)	2.8ab	0.0a	10.3a	5.0a	13.1a	5.0a					
CP, 60, 35 (1,000)	0.0a	0.0a	20.4b	2.5a	20.4b	2.5a					
MB, 60, -	0.0a	0.0a	11.8a	2.3a	11.8a	2.3a					

<sup>&</sup>lt;sup>a</sup>The value includes the percent of plants infected by *V. dahliae* and *F. oxysporum* f.sp. *lycopersici*.

Table 11 Effect of fumigation with chloropicrin (CP), applied by drip irrigation, and methyl bromide (MB) on the yield of tomato cv Principe Borghese (PB) and Vulcano (V) (Albenga, trial 6, greenhouse, 2000)

Application rate g/m <sup>2</sup> ,	$Kg/m^2$		No. fruits/m <sup>2</sup>		g/fruit	
mm water (CP µl/l)	PB	V	PB	V	PB	V
Control,-, -	2.1b <sup>a</sup>	7.1b	67c	54a	27a	128a
CP, 20, 20 (600)	2.2b	8.5ab	77b	68a	27a	123a
CP, 20, 30 (400)	2.6a	8.0ab	89a	69a	28a	113a
CP, 40, 35 (700)	2.5a	9.0a	89a	81a	26a	106a
CP, 60, 52 (700)	2.2b	8.2ab	80ab	72a	26a	109a
CP, 60, 35 (1,000)	2.2b	8.4ab	75b	74a	27a	111a
MB, 60, -	2.4ab	8.4ab	85a	68a	27a	121a

<sup>&</sup>lt;sup>a</sup> Means of the same column followed by the same letter do not differ according to Duncan's Multiple Range Test (P = 0.05).

provided by methyl bromide followed by  $40 \text{ g/m}^2$  of CP. However, it must be pointed out that only a reduction of 44% of the disease pressure was achieved. Chloropicrin, at  $20 \text{ g/m}^2$ , significantly reduced the disease pressure in

comparison to the control plots, but was less effective in comparison to the treatment with a higher dose and with methyl bromide (Table 5). When yield per area (m<sup>2</sup>) was considered, the highest production was observed in soil fumigated with MB.

#### 3.4. Trial 4 (Albenga, greenhouse drip fumigation)

The percentage of infected plants with symptoms of crown and root rot or wilt in the control plots was very high, reaching 66% and 86% at the end of the trial for cv. Principe Borghese and Vulcano, respectively (Table 7). Under such a disease pressure, MB and CP at  $40 \, \text{g/m}^2$  provided the best disease control, while the lower application rate of CP only slightly reduced the disease pressure (Table 7). In this trial, no treatment provided adequate control for the Vulcano variety. When the production per area (m²) was considered, all treatments improved the yield, particularly in the case of the cv Principe Borghese (Table 8).

<sup>&</sup>lt;sup>b</sup> Means of the same column followed by the same letter do not differ according to Duncan's Multiple Range Test (P = 0.05).

Table 12
Effect of fumigation with chloropicrin (CP), applied by drip irrigation, and methyl bromide on the survival of soil buried pathogens in trial 6 (Albenga, 2000)

Application rate g/m <sup>2</sup> , mm water (CP µl/l)	Percentage of kernels infected at the end of fumigation with											
iiiii watei (CF μi/i)	F. lycopersici		F. melonis		R. solani		S. sclerotiorum		V. dahliae			
Depth (cm)	10	20	10	20	10	20	10	20	10	20		
Control,-, -	29.0b <sup>a</sup>	20.8b	88.5c	84.8b	83.3c	51.5c	62.3b	43.3b	100.0c	96.8b		
CP, 20, 20 (600)	7.0a	10.8a	45.8b	59.5ab	12.5a	47.8bc	59.3b	11.5a	57.0b	100.0b		
CP, 20, 30 (400)	3.5a	14.8a	27.5ab	52.5ab	10.5a	24.8abc	12.5a	31.3ab	44.3ab	73.0ab		
CP, 40, 35 (700)	3.5a	8.8a	1.8a	50.0ab	9.5a	30.3abc	14.8a	39.5ab	17.3a	35.3a		
CP, 60, 52 (700)	5.5a	0.8a	13.8ab	4.8a	34.3b	12.8ab	20.5a	19.5a	42.0ab	36.8a		
CP, 60, 35 (1,000)	6.0a	2.8a	5.0ab	2.5a	21.8ab	8.0a	11.3a	7.3a	55.0b	44.3a		
MB, 60, -	0.3a	1.8a	6.0ab	36.8ab	9.8a	27.5abc	4.5a	10.0a	36.8ab	50.5a		

<sup>&</sup>lt;sup>a</sup> Means of the same column followed by the same letter do not differ according to Duncan's Multiple Range Test (P = 0.05).

## 3.5. Trial 5 (Acate, field drip fumigation)

The disease pressure was very low (diseased plants in the control plots for "Principe Borghese" and "Vulcano" were 2.5% and 4.4%, respectively, data not shown). All treatments significantly reduced the disease pressure, but did not affect the yield (data not shown). Severe infestation by TYLCV was observed. All treatments reduced the survival of R. solani, F. melonis and S. sclerotiorum at the 10 cm depth (Table 9). At the 20 cm depth, CP at  $20 \, \text{g/m}^2$  was generally less effective, particularly on S. sclerotiorum and V. dahliae. The best results in the bioassay were offered by CP applied at  $60 \, \text{g/m}^2$ , with 52 and 35 mm of water, followed by MB and CP at  $40 \, \text{g/m}^2$  (Table 9).

#### 3.6. Trial 6 (Albenga, greenhouse drip fumigation)

Disease severity was especially high in the case of the Principe Borghese variety, with severe infection of crown and root rot. The best results were obtained with CP applied at  $20 \,\mathrm{g/m^2}$  and  $30 \,\mathrm{mm}$  of water, at  $60 \,\mathrm{g/m^2}$  and  $52 \,\mathrm{mm}$  of water and with MB (Table 10). In the case of cv Vulcano, CP applied at  $40 \,\mathrm{g/m^2}$  with  $35 \,\mathrm{mm}$  of water provided the best results in terms of yield. In the case of cv Principe Borghese, CP at  $20 \,\mathrm{g/m^2}$  applied with  $30 \,\mathrm{mm}$  of water provided a very good yield. A lower production was observed when the  $20 \,\mathrm{g/m^2}$  of CP was applied with  $20 \,\mathrm{mm}$  of water or when  $60 \,\mathrm{g/m^2}$  of CP was applied with  $35 \,\mathrm{or} 52 \,\mathrm{mm}$  of water (Table 11). In the bioassay, CP at  $60 \,\mathrm{g/m^2}$  and MB at the same application rate, offered the best reduction in viability of the tested pathogens, at both depths (Table 12).

#### 4. Discussion

In our trials, in spite of the presence of severe pressure of fungal pathogens, CP applied by shank injection at rates  $\ge 30 \, \text{g/m}^2$  provided a satisfactory and consistent control of tomato diseases, without causing phytotoxicity. However, CP applied by shank injection at 30 or  $40 \, \text{g/m}^2$  was less effective than MB applied using the standard rate  $(60 \, \text{g/m}^2)$  and method.

Using different amounts of water chosen on the basis of other studies (Ajwa and Trout, 2000a, b), the concentration of CP providing the most control of fungal pathogen ranged from 400 to 700 ul/l. These results confirmed that the efficacy of CP, applied through drip irrigation systems, was affected by the amount of irrigation water more than the application rate or concentration. In the Alberga soil, the  $20 \,\mathrm{g/m^2}$ rate was not effective when applied with 17 and 20 mm of water (corresponding to 700 and 600 µl/l). The same application rate of CP applied with 30 mm of water (corresponding to 400 µl/l) was more effective both in terms of reduced pathogen viability in the bioassay and disease control, particularly in the case of the cv Principe Borghese. The application rate of 40 g/m<sup>2</sup> with 20 mm of water (1200 µl/l) did not provide satisfactory results. Better pathogen and disease control was obtained with the application of the same rate of CP in 35 mm of water (700 μl/l). These results suggest that the lower application rates of CP (20 g/m<sup>2</sup>) should be distributed with at least 30 mm of water, while higher rates (40 and 60 g/m<sup>2</sup>) should be applied with more than 35 mm of water, with up to 50 mm of water for the 60 g/m<sup>2</sup> rate, depending on the soil type and conditions and the configurations of the drip irrigation systems.

On the basis of the results obtained against the tested fungal propagules in the bioassay in Acate soil, no differences were observed between the application of  $60 \, \text{g/m}^2$  with  $35 \, \text{mm} \, (1000 \, \mu \text{l/l})$  and  $52 \, \text{mm} \, (700 \, \mu \text{l/l})$  of water. A partial improvement of CP efficacy against *S. sclerotiorum* in terms of reduced pathogen viability on in the bioassay was observed at 20 cm depth when CP was applied with 52 mm of water. No differences were observed in Albenga soils. A higher *F. radicis lycopersici* 

infestation occurred when CP was applied at  $60\,\mathrm{g/m^2}$  in 35 mm of water, but this infestation did not affect the yield.

Ajwa and Trout (2000a) applied one rate of 1,3-dichloropropene+CP (Telone C35) in three amounts of irrigation water (26, 43, and 61 mm) to sandy loam soil beds (76 cm wide) and found that the higher amount of irrigation water resulted in greater fumigant concentration in the gas phase across the soil profile. Disease control and strawberry growth and yield were also better with the higher amount of water (Ajwa and Trout, 2000b). These studies suggested that higher amounts of irrigation water used in drip fumigation increases the uniformity of fumigation and reduces volatilization losses from the soil by forming a water seal above the fumigant.

Other studies have shown that applying water and covering the soil immediately after fumigation reduces chemical volatilization from the soil (Jin and Jury, 1995). Gan et al. (1998) determined that emulsified formulations of 1,3-D applied with water resulted in the lowest amount of fumigant loss to the atmosphere compared to shank injecting 1,3-D at 20 cm depth under a PE mulch or followed by application of water to seal the soil surface. Recently, Wang et al. (2000) found that subsurface drip fumigation may be an effective method of reducing emissions as compared to conventional shank injected fumigation. Although a large amount of water might be needed for greater efficacy of drip fumigation with CP, further studies should take into account the risk of deep leaching (not yet evaluated in Italy) and water availability, particularly in Southern Italian regions, where less water than be available for agricultural uses can be reduced during the dry seasons.

Soil type and organic matter content may have influenced the efficacy of the treatment, particularly when CP was applied through shank injection. With this type of application, the same application rate of CP was more effective in Acate soil that contains more sand and less organic matter content than in the Albenga soil. Higher activity was previously observed in terms of effects on viability of fungal propagules evaluated in the bioassays (Minuto et al., 2000). However, more research is needed to determine if soil type is an important factor in the efficacy of chloropicrin in Italian soils.

The efficacy of drip fumigation was greater than shank injection, particularly in the Albenga soil for any application rate of CP, but the differences between the two soils are less evident in the bioassays than with the amount of disease control measured in the field.

#### 5. Conclusions

Chloropicrin, applied by shank injection at rates  $\ge 30 \text{ g/m}^2$ , is a viable chemical alternative to methyl

bromide soil fumigation for at least three reasons. Firstly, chloropicrin is not considered an ozone-depleting substance. Secondly, like MB, CP controls a broad spectrum of plant pathogenic fungi. Although slightly less effective than MB at the tested application rates, CP provided more than a satisfactory disease control in the presence of very high disease pressure. Lastly, CP can be applied in the same cultural and environmental conditions and situations like MB, without requiring alternative application equipment or changes in crop production practices. For agricultural sectors that still rely on soil fumigation for producing high-value crops for fresh consumption, a possible alternative to MB that provides a reliable control of fungal pathogens would be very useful, without modifying application equipment or changing production practices. In contrast to what happens with methyl bromide, the effectiveness of CP is greatly affected by the moisture content of the fungal propagules (Munnecke et al., 1982); this could explain the good efficacy of the drip application method. In order to improve the efficacy of CP to control nematode pressure, it can be mixed with specific nematocides, such as 1,3 dichloropropene (Csinos et al., 2000). However, recent research conducted in the San Joaquin Valley, CA, found that drip fumigation with CP at 300 kg/ha applied in 150 mm water controlled Pin nematode pressure in soil to a 2-m depth (Trout and Ajwa, 2000). Further research is needed to determine efficacious application rates of CP to control parasitic nematodes in soils.

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